Xenobiotic Bioavailability: Role of Intestinal Disposition

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Bioavailability and Fraction Dose Absorbed

- Bioavailability (*F*) – measurement of the rate and extent of therapeutically active drug that reaches the systemic circulation and is available at the site of action *(Shargel & Yu, 1999)*
- Fraction Dose Absorbed (f_a – fraction of oral) dose that traverses the intestine intact

Intestinal Disposition

- Intestinal permeability, metabolism,solubility stability and dissolution of a xenobiotic
- Inhibition of membrane transporters and/or metabolizing enzymes
- Modulation of the expression of membrane transporters and/or metabolizing enzymes

Factors Affecting Oral Bioavailability

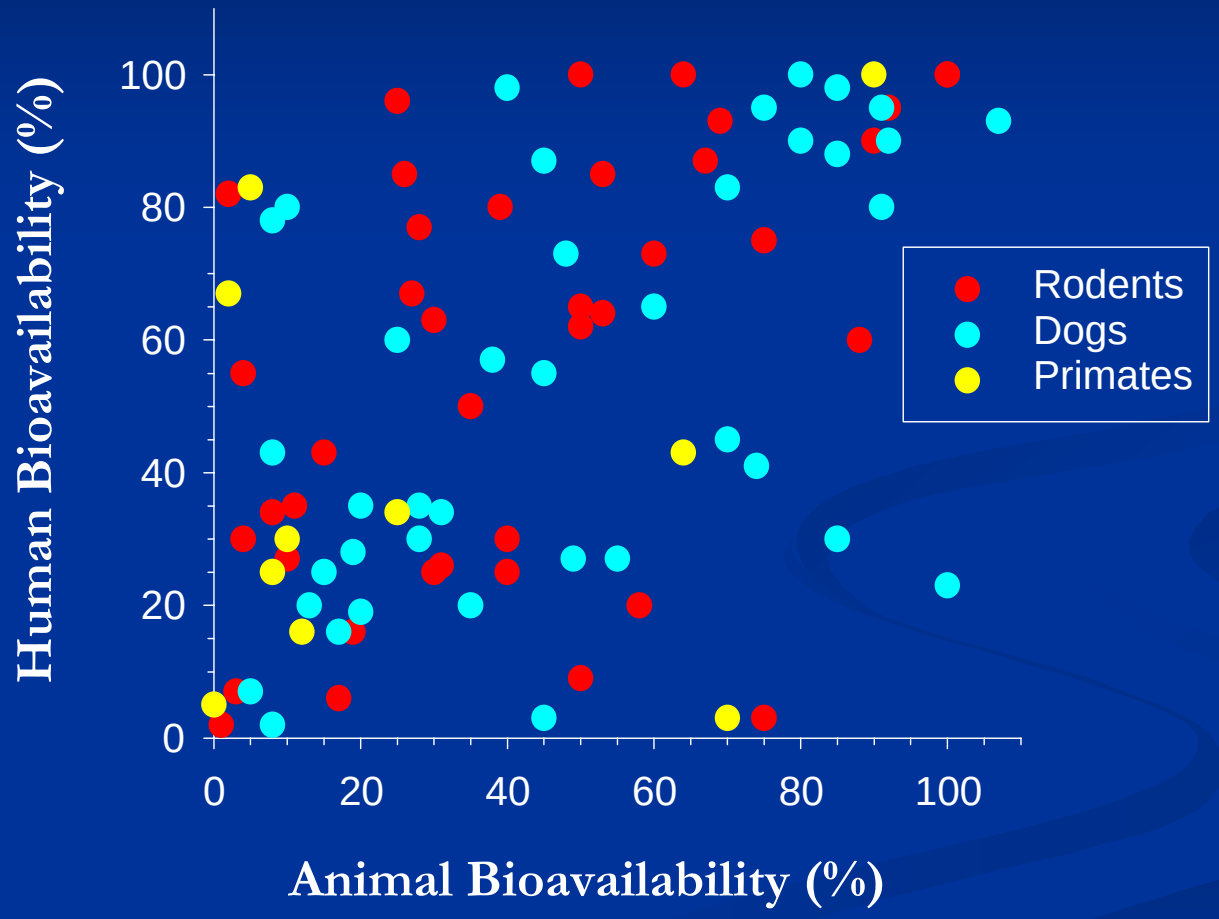
Physicochemical

- Solubility
- Ionization
- Dissolution Rate
- Chemical Stability
- Diffusion (intra-lumenal)

Physiological

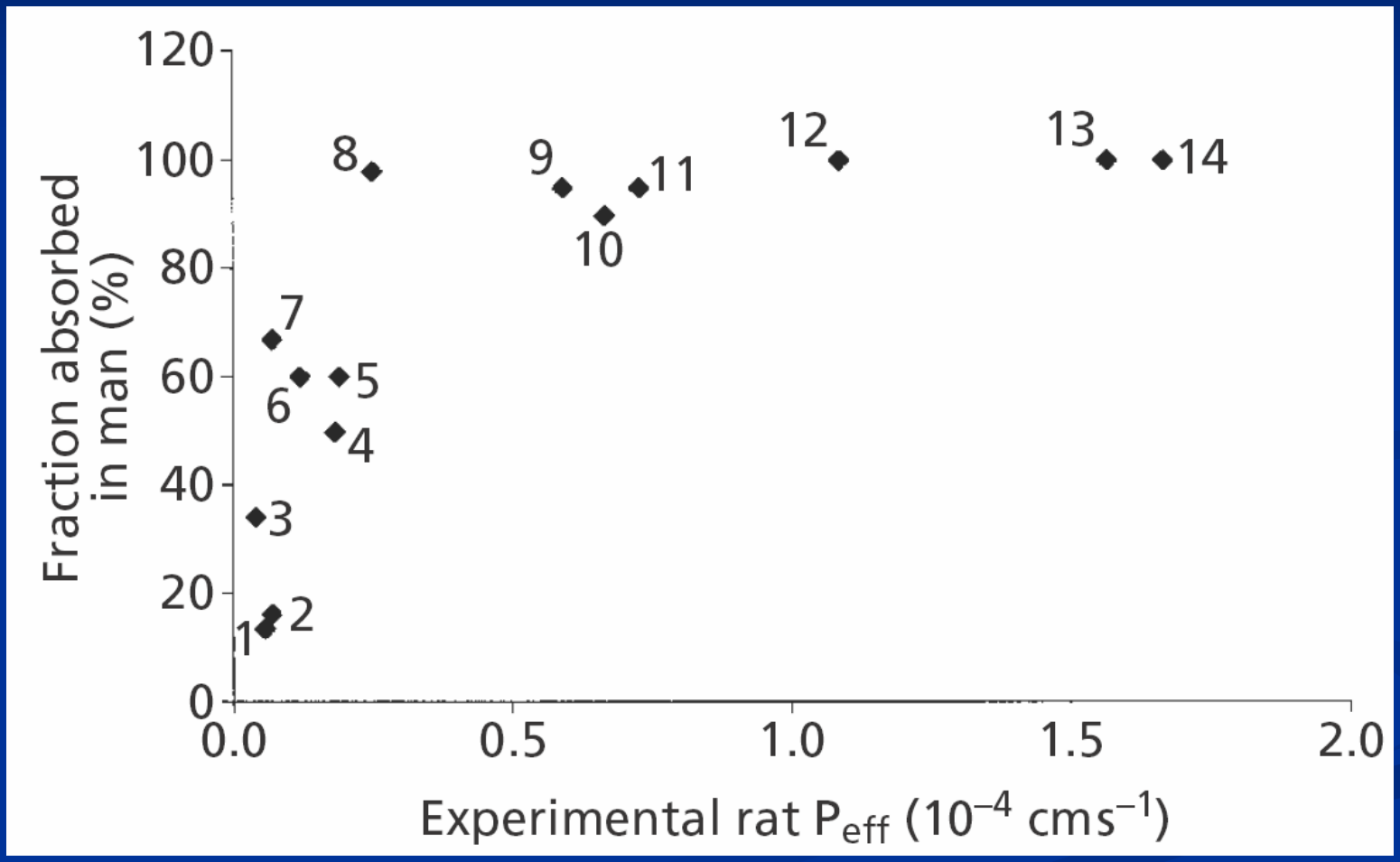
- GI Transit
- Bile Secretion
- Transport Mechanisms
- Metabolism
- Regional Effects
- Polymorphism of Transporters/Enzymes

Oral Bioavailability Comparison



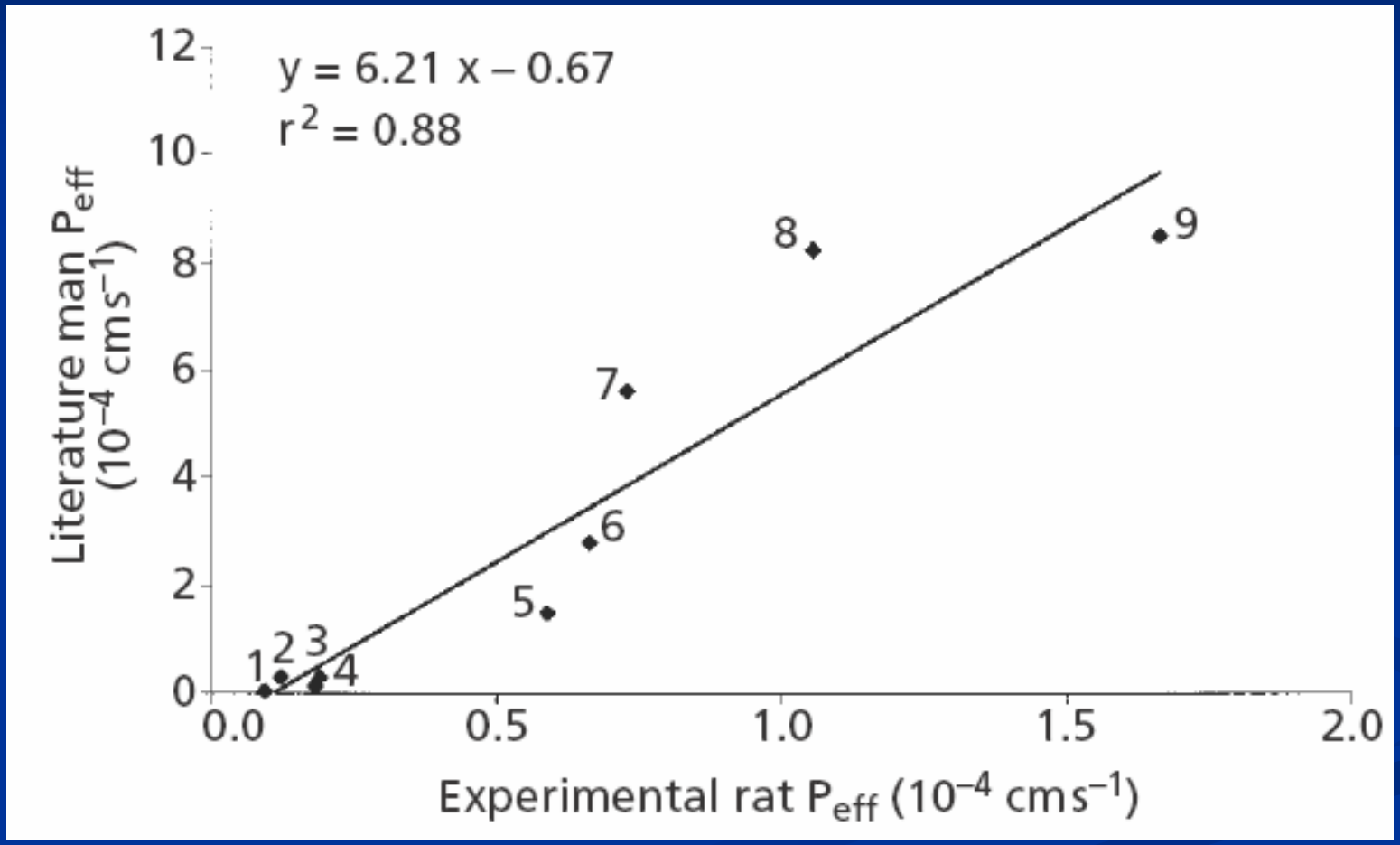
Adapted from: W.K. Sietsema, *Int. J. Clin. Pharmocol. Ther. Toxicol.*, **27**:179-211 (1989)
G. Grass

Fa – Permeability Comparison



From: L. Salphati, et. al., *J. Pharmacy Pharmocol.*, **53:**1007-1013 (2001)

Permeability – Permeability Comparison



From: L. Salphati, et. al., *J. Pharmacy Pharmocol.*, **53:**1007-1013 (2001)

Intestinal Transport and Metabolism

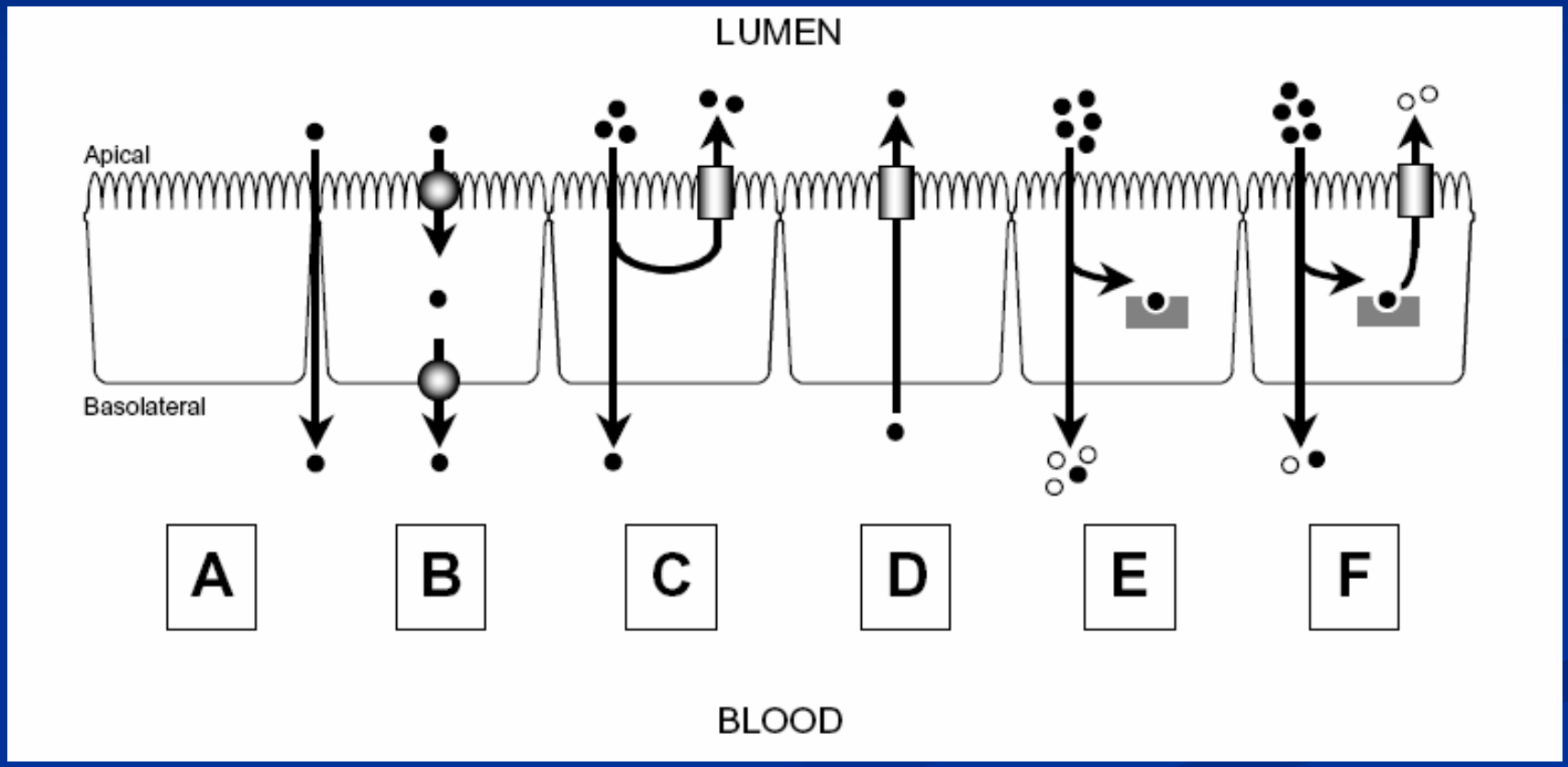


Figure from *Eur J Pharm Sci* **21**: 25, 2004

Proteins Involved in Intestinal Disposition

- Influx Transporters
  - Peptide, bile acid, nucleoside, amino acid, etc.
- Efflux Transporters
  - P-gp, MRP2, BCRP, etc.
- Metabolizing Enzymes
  - Phase I –– CYP isoforms (primarily 3A4, 2D6,
  - Phase II –– GSTs, UGTs sulfotransferases
- Nuclear Hormone Receptors

  CAR, PXR, PPAR, RXR etc.

Intestinal Disposition

- Permeability
- Metabolism

Also

- Inhibition
- Induction

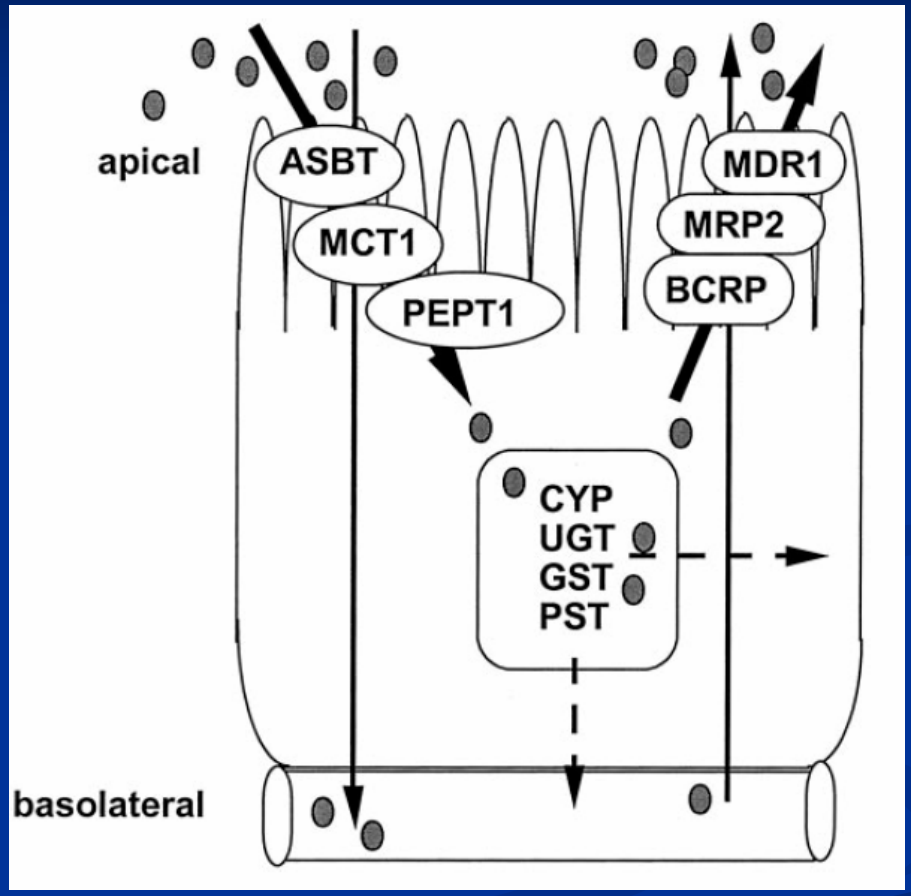
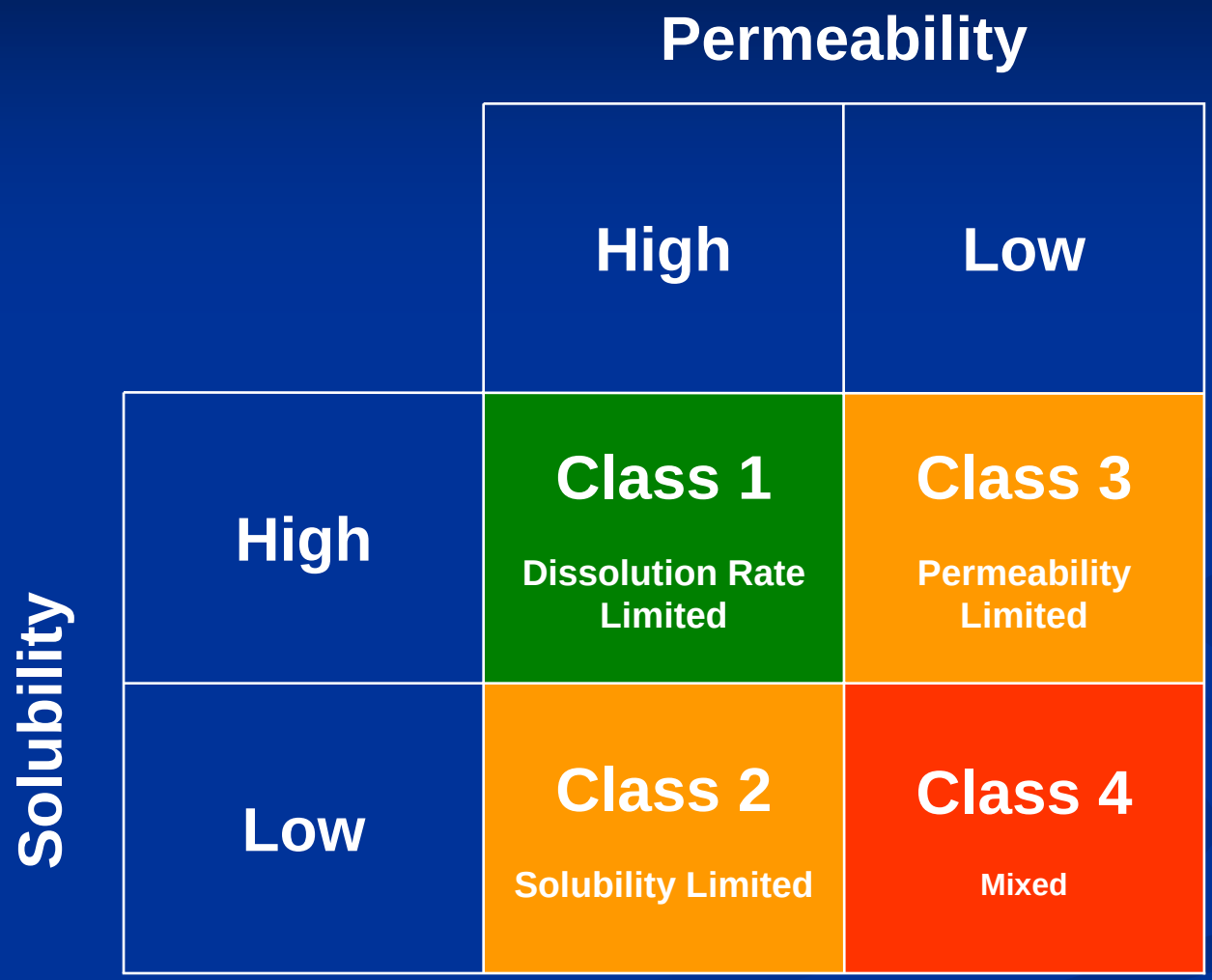
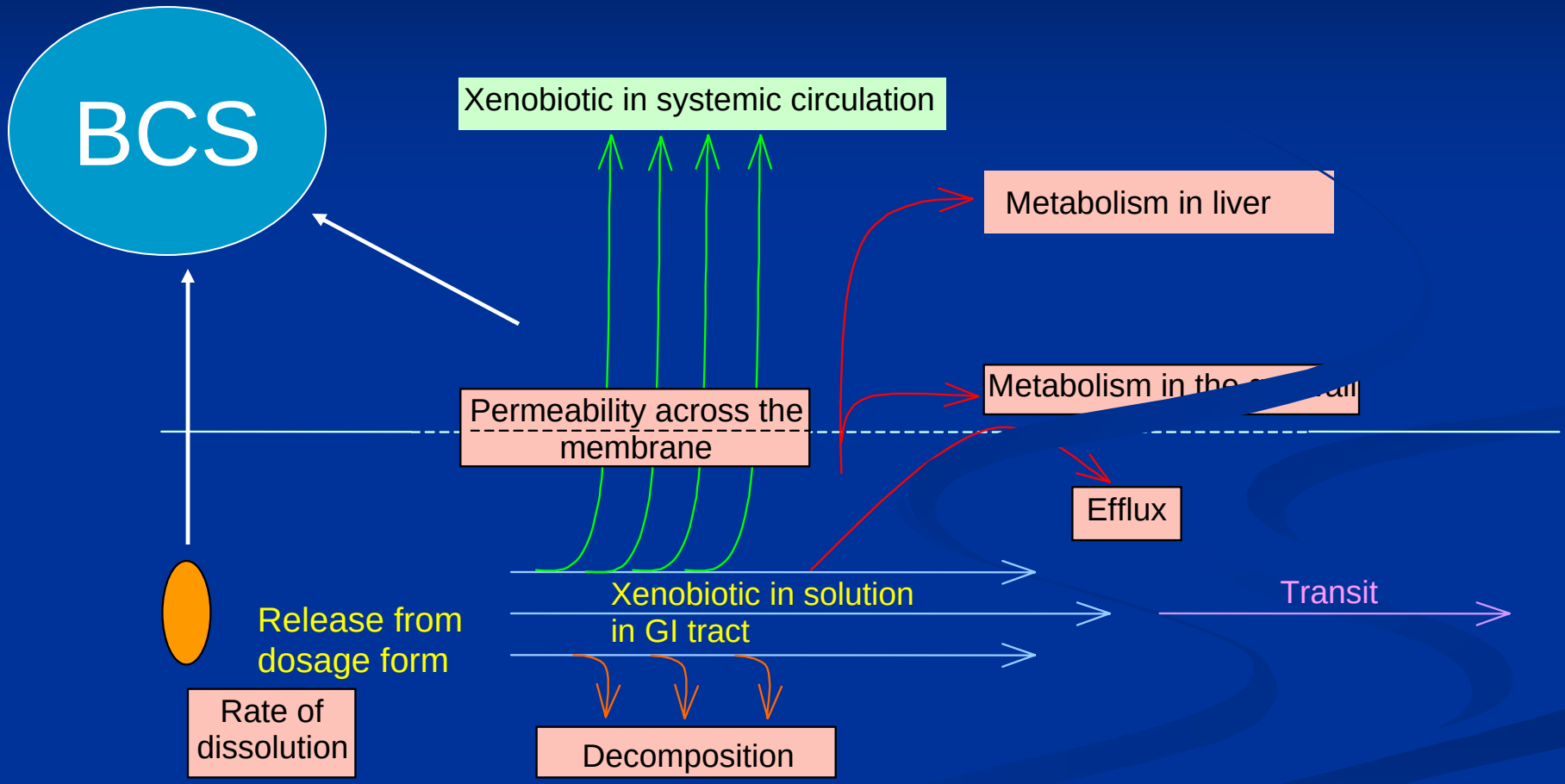


figure from *Drug Metab Dispos* **31**: 1507, 2003

Biopharmaceutical Classification System



Factors affecting rate and extent of oral absorption



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Clinical Intestinal Metabolism Drug Interactions

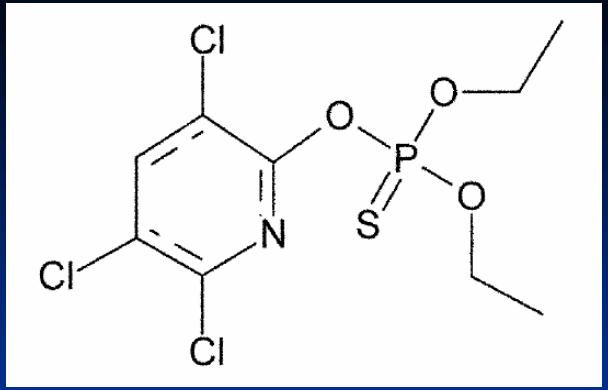
Drug	Interacting agent	Effect on relative bioavailability	Inhibition or induction	Clinical significance
Immunosuppressants				
Cyclosporin	Ketoconazole	↑ × 2.5	Inhibition	High cyclosporin concentrations, enabling a reduction in dosage
	Erythromycin	↑ × 2.0	Inhibition	
	Grapefruit juice	↑ × 0.6	Inhibition	
Tacrolimus	Rifampicin	↓ × 2.7	Induction	Therapeutic failure: transplant rejection
	Ketoconazole	↑ × 2	Inhibition	High concentrations: dosage reduction
Antivirals				
Saquinavir	Grapefruit juice	↑ × 2	Inhibition	High concentrations: dosage reduction
Indinavir	St John's wort	↓ × 2	Induction	Therapeutic failure
Cardiac				
Nifedipine	Rifampicin	↓ × 8	Induction	Therapeutic failure
	Grapefruit juice	↑ × 2.8	Inhibition	Potential toxicity
Felodipine	Erythromycin	↑ × 2.5	Inhibition	Increased toxicity: low blood pressure
	Grapefruit juice	↑ × 1-2	Inhibition	
	Itraconazole	↑ × 6	Inhibition	
Other				
Simvastatin	Itraconazole	↑ × 5	Inhibition	Potential for increased skeletal muscle pain
	Grapefruit juice	↑ × 16	Inhibition	
	Erythromycin	↑ × 4	Inhibition	
Lovastatin	Grapefruit juice	↑ × 15	Inhibition	Potential for increased skeletal muscle pain
	Cyclosporin	↑ × 20	Inhibition	
	Itraconazole	↑ × 20	Inhibition	

(adapted from:
Doherty, M.M. and Charman, W.N., *Clin Pharmacokinet*, **41**:235-253, 2002)

Preclinical Methods for Intestinal Disposition

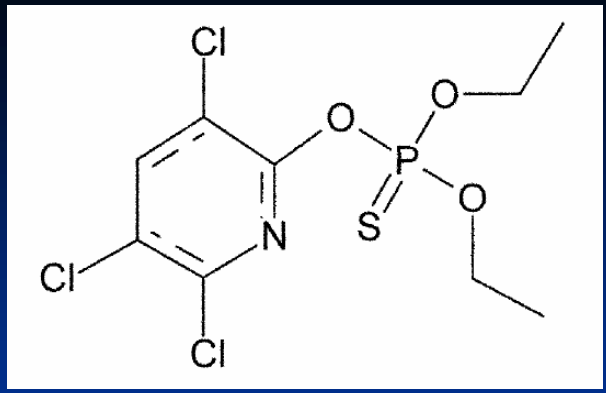
- Intestinal permeability studies
  - Perfusion
  - Diffusion chamber (excised tissue or cultured cells)
  - Everted gut sac
  - PAMPA
- Intestinal metabolism
  - Perfusion
  - Microsomes
- Oral PK studies
  - P-gp, CYP inhibitors
  - Knockout animals

Chlorpyrifos



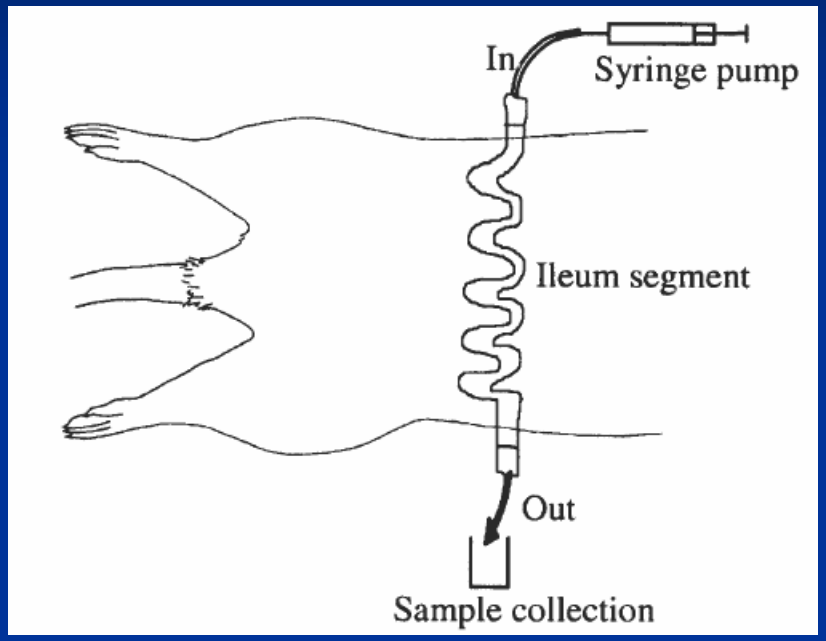
- Organophosphate pesticide
- Potential exposure routes
- Limited human bioavailability studies
- Goals
  - Determine intestinal permeability as a function of region and concentration
  - Determine effect of chlorpyrifos on expression and function of membrane transporters

Chlorpyrifos



- Single-pass Intestinal Perfusion (SPIP)
  - Regional permeability as a function of concentration
- Exposure studies in Caco-2 cells
  - Competitive PCR assay for MDR1
  - Effect on membrane efflux function

Single Pass Intestinal Perfusion



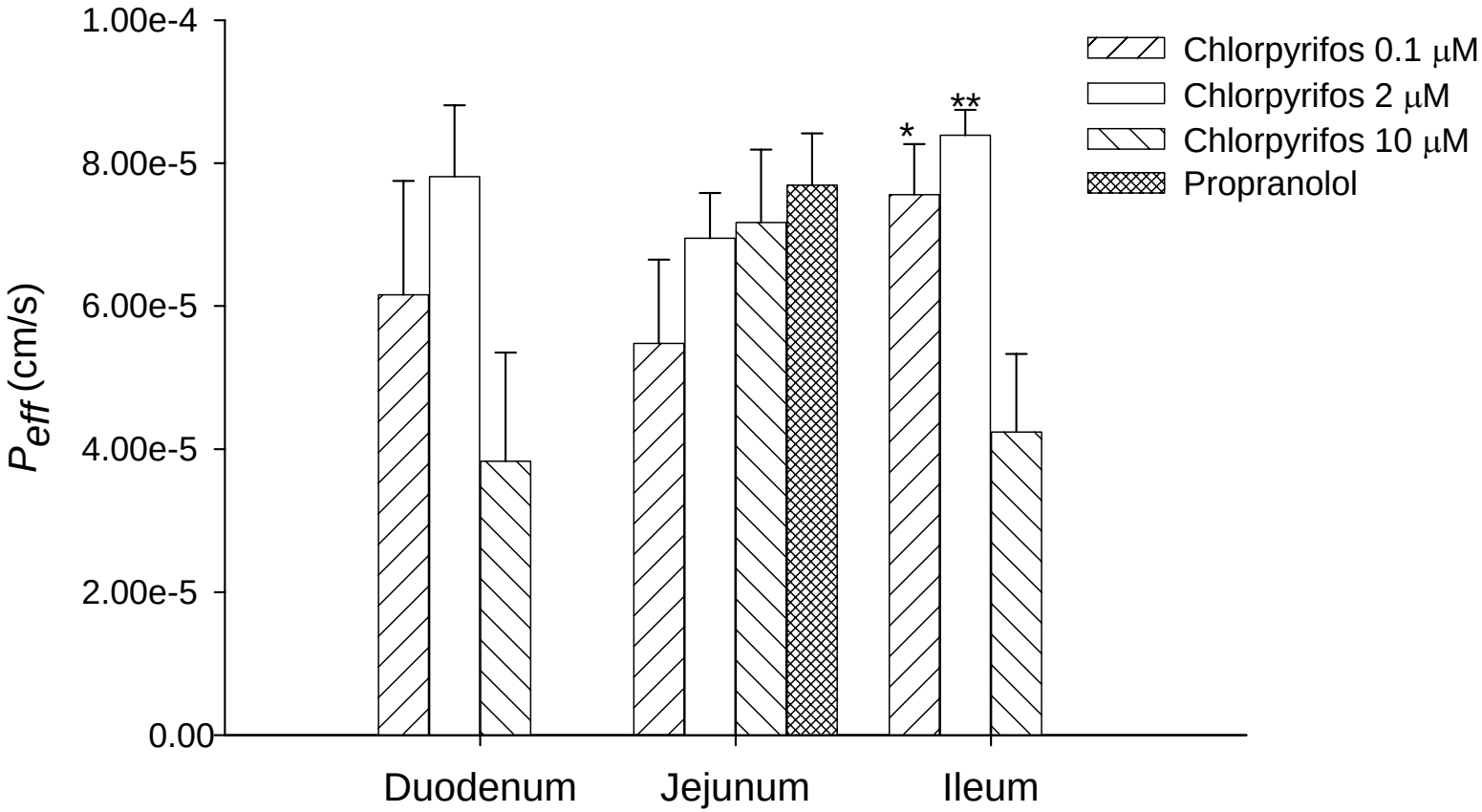
- Permeability determined by loss from perfusate

$$P_{eff} = \frac{-Q}{2\pi r l} \ln\left(\frac{C_{out}}{C_{in}}\right)$$

- Correct for adsorption, stability, accumulation

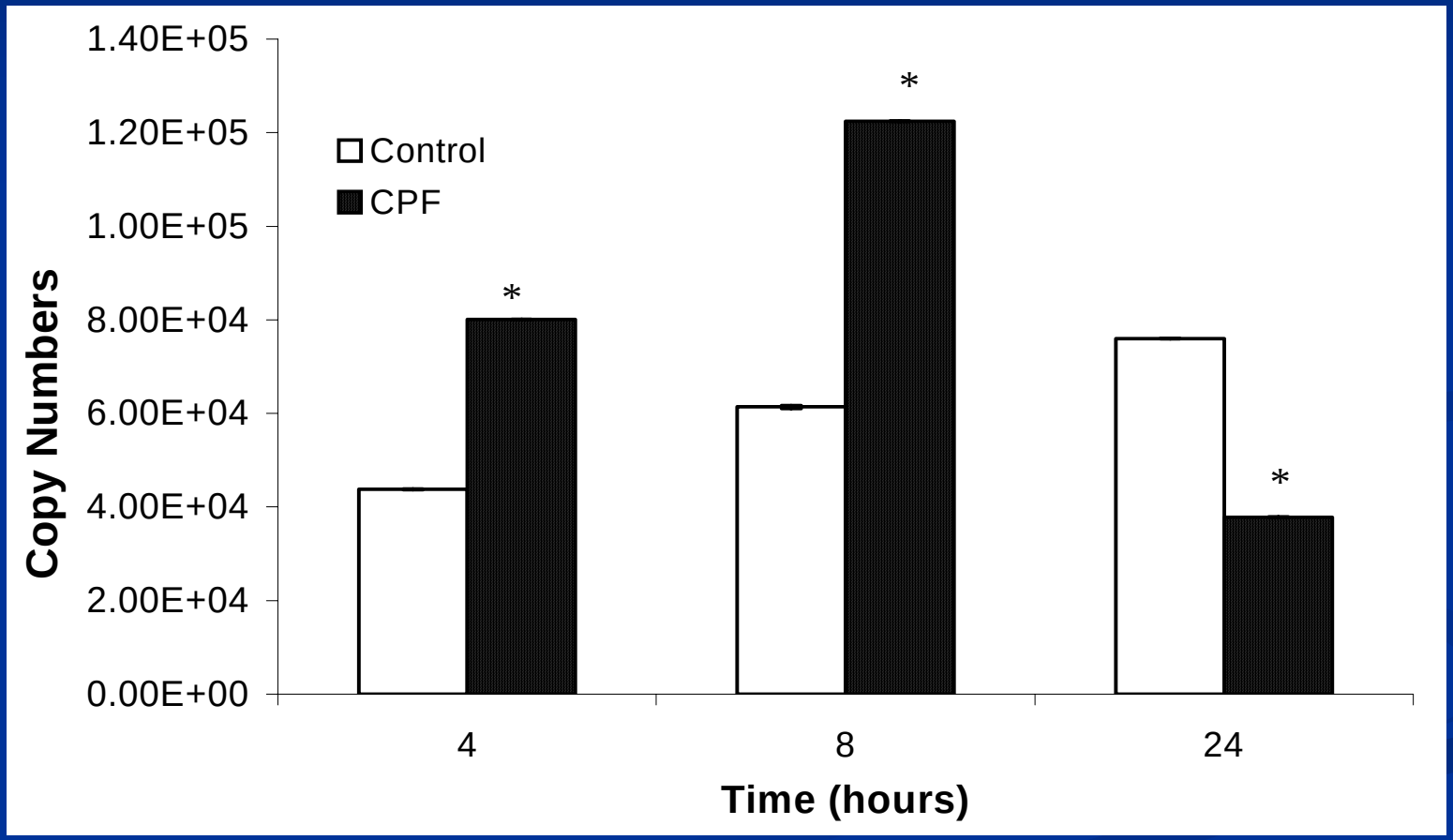
Figure from: Salphati, et al, JPP 2001, **53**: 1007–1013

Results – Permeability



T.J. Cook and S.S. Shenoy, *Toxicology*, **184**:125-133, 2003

Results – Effect of CPF on MDR1 Expression in Caco-2 Cells



S. Agarwala, W. Chen and T.J. Cook, *Toxicol. In Vitro*, **18**:403-409 (2004)

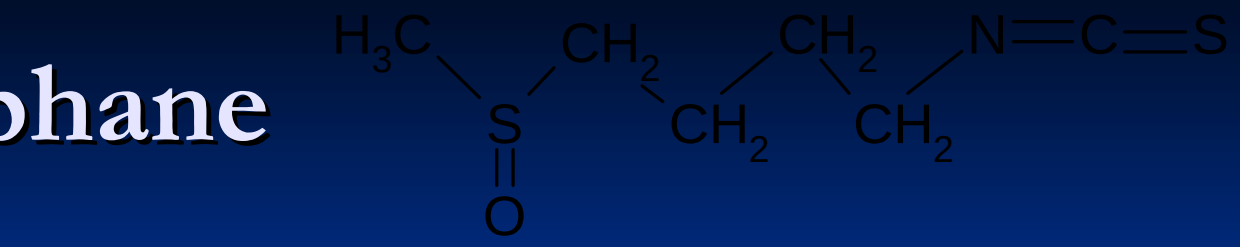
Results

Effect of CPF on Efflux Function in Caco-2 Cells

	VL	*VC*	*VH*
Control	2.87	3.40	4.44
8 hr CPF pre-incubation	3.65	4.18	5.01
Increase	27%	23%	13%

S. Agarwala, W. Chen and T.J. Cook, *Toxicol. In Vitro*, **18**:403-409 (2004)

Sulforaphane



- Isothiocyanate from cruciferous vegetables
- Potential chemopreventive agent
- Mechanism of action
  - Induction of Phase II metabolizing enzymes and efflux transporters, e.g., MRP2
- Goal: Determine intestinal disposition and effect of SFN on expression of Phase II enzymes and MRP2 in intestine

SPIP-MV

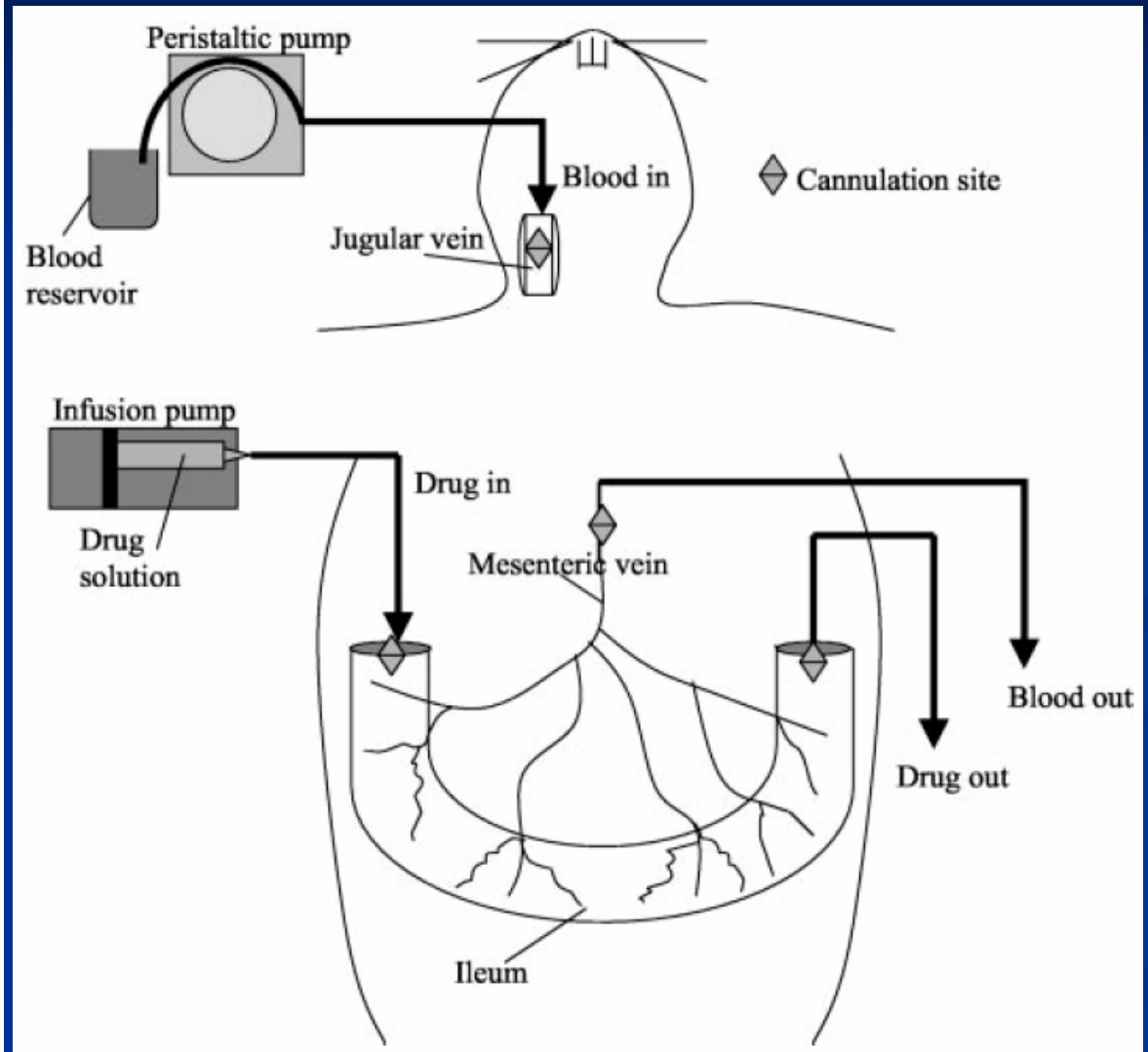
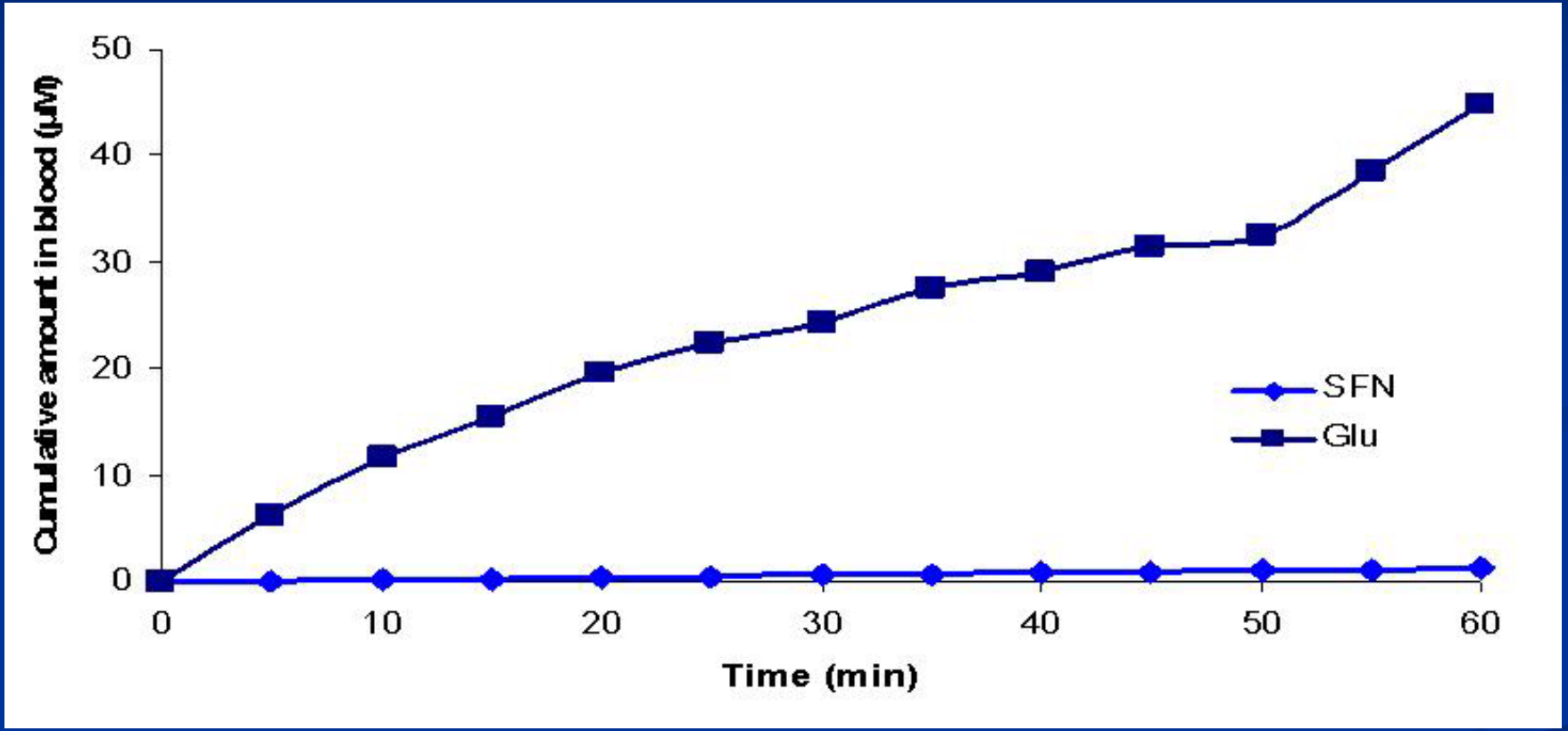


Fig. 1. Illustration of the experimental setup for single-pass intestinal perfusion with mesenteric cannulation and continuous infusion of blood through the jugular vein.

- Permeability Determination
  - Lumenal
  - Blood
- Bioanalytical

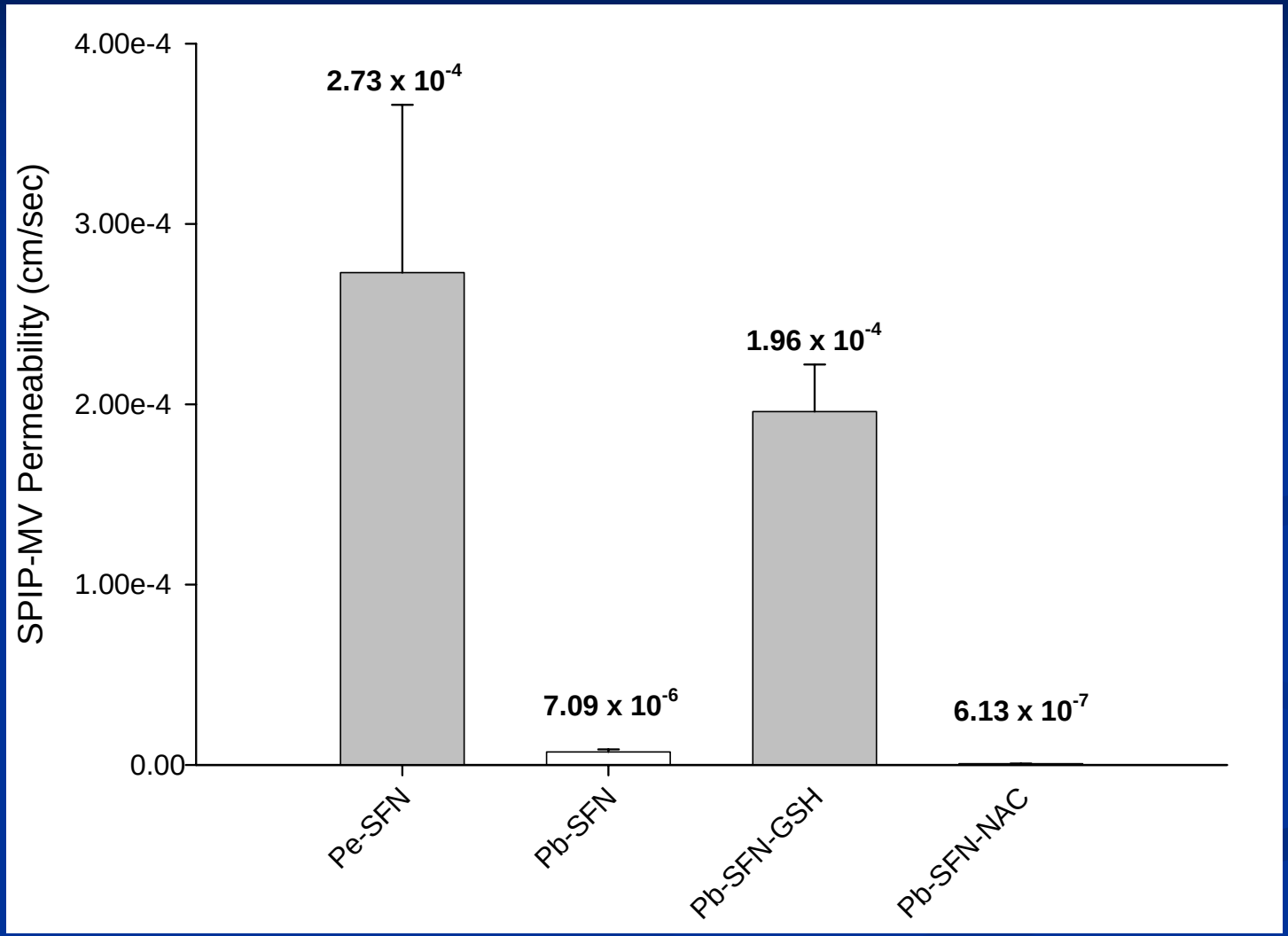
Figure from Cummings, et al, *JPET*, **305**:306, 2003.

SFN and SFN-GLU in Mesenteric Blood



Agrawal, S., Tsao, Y., Hu, P., and Cook, T.J., Intestinal Disposition of Sulforaphane, In Preparation, 2005.

Permeability of SFN and Metabolites



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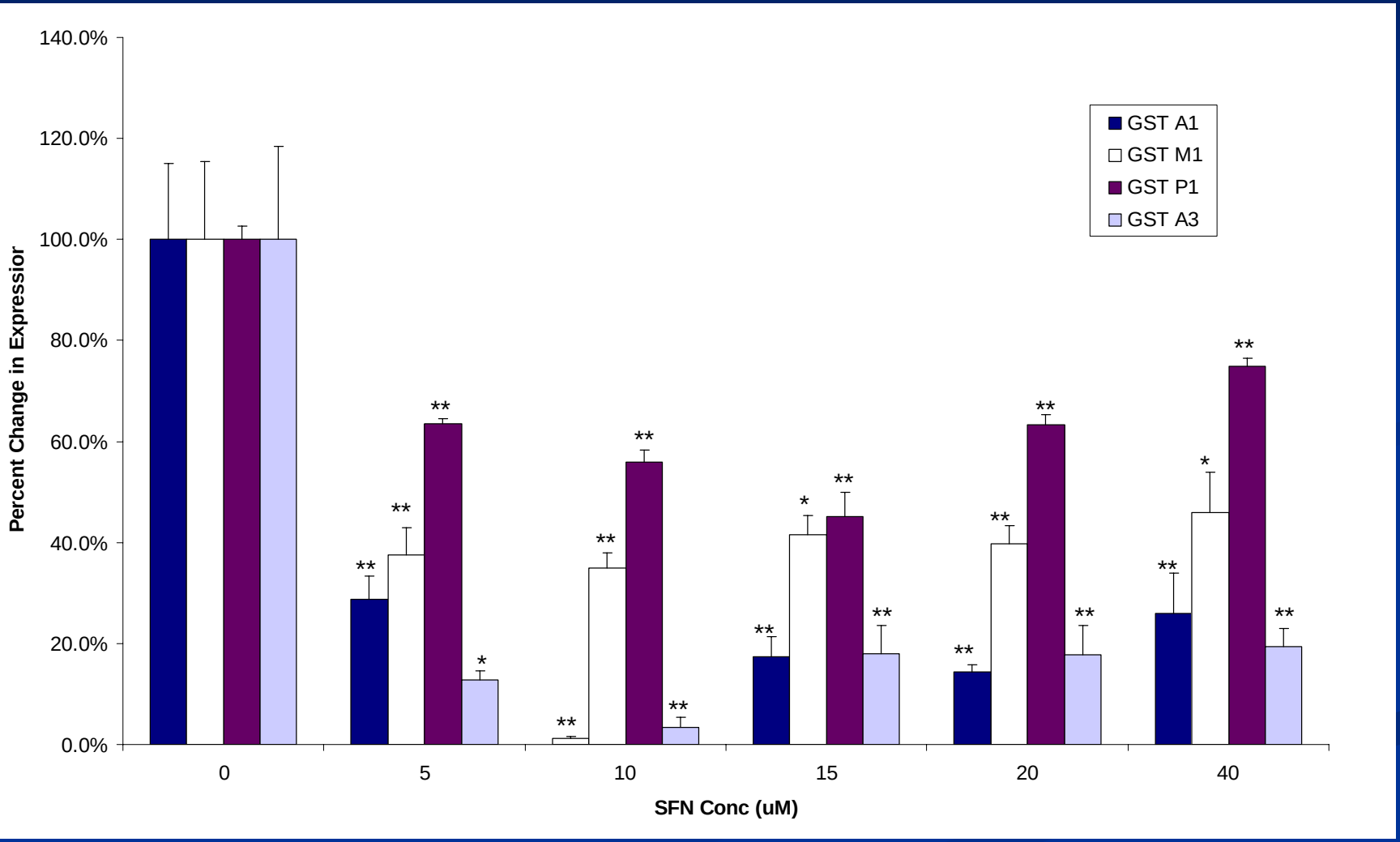
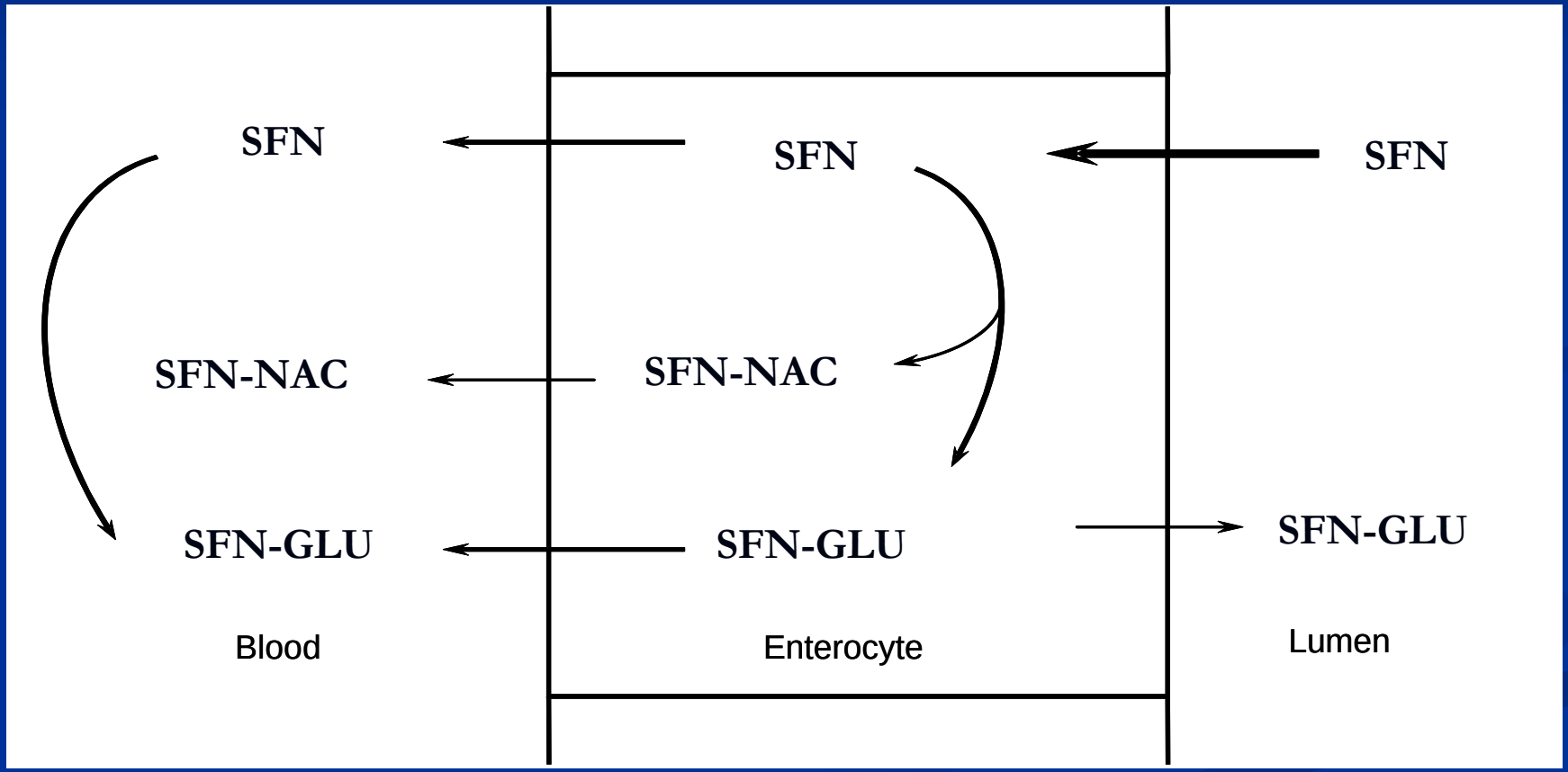
Effect of SFN on GST Expression in Rat Ileum



Tsao, Y., Hu, P., and Cook, T.J., AACR Frontiers in Cancer Prevention Research, #A133, 2004.

Model of SFN Intestinal Disposition



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Relevance

- Depends on:
  - Metabolic pathways
  - Therapeutic index of drug
  - Toxicity of xenobiotic
  - Variability in intestinal metabolism
- Xenobiotic – Drug Interactions
  - Induction of expression
  - Relative affinity for transporter/enzyme
  - Concentration, etc
  - Exposure

Summary

- Intestinal disposition is critical for the bioavailability of orally administered compounds *(but may not be the limiting factor)*
- Interactions with transporters/enzymes (modulation of expression and/or function) should be considered
- Dietary factors (e.g., grapefruit juice) can contribute to variability in oral drug bioavailability
- "Baseline" expression of patients may change based on dietary factors
- Potential contribution of unidentified transporters/enzyme isoforms

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